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EXAMINER	
BAUSCH, SARAE L	
ART UNIT	PAPER NUMBER
1634	
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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/501,040	BERLIN, KURT	
	Examiner	Art Unit	
	Sarae Bausch	1634	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *25 April 0207*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-43* is/are pending in the application.

4a) Of the above claim(s) *42 and 43* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-41* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date *01/07*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

1. This action is in response to applicants correspondence mailed 09/04/2007. The amendment to the drawings mailed 09/04/2007 has been entered

Election/Restrictions

2. Applicant's election of group I, claims 1-41 in the reply filed on 04/25/2007 is acknowledged. Because applicant did not distinctly and specifically point out the supposed errors in the restriction requirement, the election has been treated as an election without traverse (MPEP § 818.03(a)).

3. Claims 42-43 are withdrawn from further consideration pursuant to 37 CFR 1.142(b) as being drawn to a nonelected invention, there being no allowable generic or linking claim. Election was made **without** traverse in the reply filed on 04/25/2007.

Drawings

4. The drawings are acceptable.

Claim Rejections - 35 USC § 112- Second Paragraph

5. The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

6. Claims 4-5, 14-15, 18-20, 22, 30-39 rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claims 4-5 recite "the genomic DNA sample (or the DNA to be investigated)" however this recitation renders the claim indefinite. It is unclear if the parenthetical phrase is intended as a required limitation of the claim.

Claims 14-15 and 30, recites the limitation "the background DNA" in line 2 of claims 14 and claim 15 and line 3 of claim 30. There is insufficient antecedent basis for this limitation in the claim. Claims 14-15 and 30 depend from claim 1. Claim 1 does not recite a background DNA.

Claims 18 recites "bisulfite (= disulfite, hydrogen sulfite)" however this recitation renders the claim vague and indefinite. It is not clear if applicant intends for the claimed method to be limited to the recited hydrogen sulfite and disulfite. Claims 19-20 depend from claim 18 and are therefore vague and indefinite for the reasons above.

Claim 22 recites "chromatography (e. g. HPLC)" however this recitation renders the claim vague and indefinite. It is not clear if applicant intends for the claimed method to be limited to HPLC.

Claim 31 recites "two blocker oligonucleotides (or blocker PNAs, generally blocker molecules)" however this recitation renders the claim vague and indefinite. It is not clear if the applicant intends for the claimed method to be limited to the recited blocker PNA and generally blocker molecules. Claims 32-36 depend from claim 31 and are therefore vague and indefinite for the reasons above.

Claim 37 recites "the probe oligonucleotides (probes)" however this recitation renders the claim vague and indefinite. It is unclear if the parenthetical phrase is intended as a required limitation of the claim. Claims 38-39 depend from claim 37 and are therefore vague and indefinite for the reasons above.

Claim Rejections - 35 USC § 102

7. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

8. Claims 1, 7- 9, 18-20, 21-29, and 41 are rejected under 35 U.S.C. 102(b) as being anticipated by Olek (US Patent 6214556)

With regard to claim 1, Olek et al. teach a genomic DNA sample is converted by treatment with a bisulfite solution to convert cytosine to uracil followed by amplification of the treated genomic DNA by short oligonucleotides complementary to adaptor oligonucleotides that have been ligated to the end of the DNA (chemically treated DNA is amplified by at least 2 pairs of complementary probes as well as a ligase) (see column 6, lines 58-67). Olek et al. teach detection of the amplificates by hybridization to draw conclusions regarding the phenotype of the analyzed cell material (methylation status) (see column 7, lines 4-11 and 19-25).

With regard to claim 3, Olek et al. teach the methylation status of the DNA is concluded by hybridization (concluded from analysis of additional positions of the amplification) (see column 7, lines 19-25).

With regard to claim 7, Olek et al. teach the primers and adaptors bear a phosphate group at the 5' end (see column 16, lines 47-51)

With regard to claims 18-20, Olek et al. teach treating extracted DNA to bisulfite treatment in sheared (claim 20) and single stranded DNA form (see column 11, lines 20-25) and teach the inclusion of the bisulfite reaction in an agarose matrix (claim 19)(see column 12, lines 4-9).

With regard to claim 21 and 41, Olek et al. teach analysis of DNA hybridization on a DNA chip (multiplex) (See column 17, lines 28-50).

With regard to claim 22, 25, 28-29, Olek et al. teach analysis by mass spectroscopy (see column 21, lines 65-67). Olek et al. teach phosphothioate analogs of amplificates for mass spectroscopy detection.

With regard to claim 23, Olek et al. teach sequence analysis by hybridization (see column 6, lines 58-67).

With regard to claim 24, Olek et al. teach determination of shared genetic and or biochemical features of tumor cells and capable of detecting changes in genetic expression of tumor cells(conclusion made on presence of disease from methylation status at different CpG positions) (See column 10, lines 30-56).

With regard to claim 8-9 and 25-26, Olek et al. teach labeling the amplificate by an incorporated nucleotide analog that is fluorescent (see column 20, lines 23-47).

9. Claims 1-13, 18, 20, 22, 24-26, and 37-41 rejected under 35 U.S.C. 102(b) as being anticipated by Nazarenko et al. (US Patent 5866336).

Nazarenko et al. teach a method of methylation analysis using oligonucleotides that are detectably labeled with molecular energy transfer labels which provide a rapid, sensitive, and reliable method for detecting amplification products that greatly decreases the possibility of carryover contamination with amplification products and that is adaptable to many method for amplification of nucleic acid sequences (see column 2, lines 42-52).

With regard to claim 1, 6, 18 and 37-40, Nazarenko et al. teach contacting a sample containing nucleic acids with bisulfite (claim 18) to convert unmethylated cytosines to uracil and conducting an amplification reaction in the presence of primers specific for selected target sequences (see column 32, lines 3-16). Nazarenko et al. teach a triamplificate reaction which uses of a third oligonucleotide, blocker oligonucleotide that is ligated to the extended hairpin primer (see figure 6). Nazarenko et al. teach the two primers are complementary to the target strand and a third oligonucleotide, blocker, is partially complementary to one of two extending primers. Nazernko et al. teach the triamplificaiton utilized two thermostable enzymes, DNA polymerase and DNA ligase (claim 38 and 39). Nazarenko et al. teach during the repeated steps of polymerization and ligation, one of the extended primers is ligated to the blocker. Nazarenko et al. teach the use of hairpin primers in the amplification reaction, during the first cycle the hairpin primer will be extended and ligated to the blocker probe, during the second cycle the extended hairpin primer will become a template for the second primer (see column 26, lines 36-43).

With regard to claim 2, Nazarenko et al. teach the DNA investigated is preferred over background DNA (methylated, unmethylated, and untreated DNA) (see example , column,

With regard to claim 4-5, Nazarenko et al teach primers specific for both methylated and unmethylated nucleic acids (see column 32, lines 8-10).

With regard to claim 7, Nazarenko et al. teach primers that have a phosphate group at the 5'end (see figure 6).

With regard to claim 8-11 and 22, Nazarenko et al. teach oligonucleotides with fluorescent labels that are a donor and acceptor moiety (see column 17, lines 15-22). Nazarenko et al. teach the donor and acceptor moieties will be at a distance wherein the emission of the donor moiety are absorbed by the acceptor moiety (see column 18, lines 43-59).

With regard to claim 12-13, Nazarenko et al. teach the fluorescence is either quenched or increased by the donor and acceptor moieties (see column 18, lines 48-59).

With regard to claim 24, Nazarenko et al. teach the method is used for diagnosis of an infectious disease (see column 19, lines 15-20).

With regard to claim 25-26, Nazarenko et al. teach the amplification product generates a FRET signal (labeled, with fluorophore) (see column 29, lines 35-37)

With regard to claim 27, Nazarenko et al. teach the labels with radioactive labels (see column 18, line 62).

With regard to claim 41, Nazarenko et al. teach the amplification reaction can be repeated for the same sample with different sets of primer that amplify wild type or mutated versions (see column 19, lines 25-33, column 30, lines 63-67) (multiplex reaction)

Claim Rejections - 35 USC § 103

10. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

11. Claims 1-7, 9, 14-18, 20, 22-25, 27, and 37-41 are rejected under 35 U.S.C. 103(a) as being unpatentable over Herman et al. (PNAS 1996, vol. 93, pp. 9821-9826) in view of Barany et al. (US Patent 5494810).

Herman et al. teach a method of determining methylation status of CpG sites in nucleic acids. Herman et al. teach a method that is specific and sensitive for methylation of any bock of CpG sites in a CpG island. Herman et al. teach genomic DNA was modified with bisulfite which converts all unmethylated cytosines to uracils (see bisulfite modification). Herman et al. teach PCR amplification of the bisulfite modified DNA with primer pairs (at least 2 pairs of essentially complementary probe oligonucleotides) followed by restriction digestion of the amplificates and/or gel electrophoresis for determination of the presence of amplificate (see pg. 9821-2, PCR amplification and Restriction Analysis, figure 2). Herman however does not teach the use of a ligase.

With regard to claim 3, Herman et al. teach the methylation status in the DNA is determined by analysis of additional positions in the amplificate by restriction digestion of the amplificates followed by gel electrophoresis (see figure 2 and pg. 9821-2, PCR amplification and Restriction analysis).

With regard to claim 4-5, Herman et al. teach specific amplification of methylated and unmethylated alleles. Herman et al. teach methylation specific primers and primers designed for unmethylated alleles (see pg. 9823, 1st column, 1st two para., pg. 9824, 1st column, 1st full para.) (probe that hybridize to template of genomic DNA that were methylated and probes designed to hybridize to unmethylated template).

With regard to claim 7, Herman et al. teach amplification primers which have a phosphate group at the 5'end (see pg. 9822, table 1 and PCR amplification).

With regard to claims 2, 14-15, Herman et al. teach the level of sensitivity of methylation sensitive PCR is 1 :1000, methylated DNA to background DNA concentration (see pg. 9826, 1st column, 1st para. and figure 2)

With regard to claim 17, Herman et al. teach genomic DNA is obtained from tissue samples (see pg. 9821, 2nd column, last para).

With regard to claim 18 and 20, Herman et al. teach treating genomic DNA with sodium bisulfite and NaOH (denatures DNA duplex) (see pg. 9822, 1st column, 1st full para).

With regard to claim 22, Herman et al. teach analysis of amplificates by gel electrophoresis (see figure 2).

With regard to claim 24, Herman et al. teach determining the methylation status of p16 in human cancer cells (see pg. 9823, 1st column, 2nd last para. and pg. 9824, 1st column, last para).

With regard to claim 39, Herman et al. teach use of Taq polymerase (see pg. 9822, 2nd column, 1st para.)

However, Herman et al. does not teach the use of a ligase, detectable label on the probe, or sequencing analysis of the amplificate.

Barany et al. teach ligase-mediated DNA amplification to detect genetic diseases. Barany et al. teach the ligase mediated DNA amplification method allows for amplification of DNA from 200 copies in 3 hrs using 30 cycles. Barany et al. teach the LCR assay can be used to detect genetic disease such as cancer (See column 46 line 25-27). Barany et al. teach denatured DNA is hybridized to two oligonucleotide probes that are immediately adjacent each other, the 3' end of one is adjacent to the 5'end with ligase allowing for a the thermophilic ligase (claim 40) to efficiently link correctly base paired oligonucleotide and give zero background ligation in the presence of a mismatched sequence (see column 29, lines 25-67 con't to column 30, lines 1-10). Barany et al. teach that the ligation product of one cycle become the target for the next cycle of ligation (see column 30, lines 14-18 and figure 2) (claim 6). Barany et al. teach oligonucleotides were labeled with ^{32}P (see column 31, lines 44-55 and column 32, lines 20-34)(claim 9, 25, and 27). Barany et al. teach detection of ligation product by hybridization (claim 23) (see column 41, lines 23-27 and column 46, lines 18-25). Barany et al. teach analysis of specific sequences from a clinical sample can be analyzed and teach the clinical sample can be blood, serum or urine (see column 38, lines 43-60) (claim 16). Barany et al. teach PCR amplification to obtain DNA targets followed by ligase chain reaction (see column 43, lines 14-20 and lines 60-67, table column 42) (claim 37-39). Barany et al. teach tandem ligation assay and detection of different genes in a single assay (multiplexing) (see column 45, lines 34-42) (claim 41).

Therefore, it would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made to improve the method of detecting methylation of a target DNA using amplification by PCR as taught by Herman et al. to include ligase chain reaction as taught by Barany et al. to make the method of Herman et al. more versatile. The ordinary artisan

would have been motivated to improve the method of analyzing the methylation pattern of the DNA by Herman et al. with the method of the ligase chain reaction by Barany et al. because Barany et al. teaches that ligase chain reaction can efficiently link correctly base paired oligonucleotide and give zero background ligation in the presence of a mismatched sequence. The ordinary artisan would have had a reasonable expectation of success that the use of ligase chain reaction could be used in the method of Herman et al. because both Herman and Barany teach amplification of a target nucleic acid to detect cancer. Furthermore, because both Herman and Barany teach analysis of target nucleic acid by amplification to detect diseases, specifically cancer, it would have been obvious to one skilled in the art to substitute one method, the method of PCR amplification of the genomic DNA sample as taught by Herman, for the method of the ligase chain reaction of the genomic DNA sample as taught by Barany in order to achieve the predictable result of detecting target gene sequences for analysis of cancer detection.

12. Claims 14-15 and 30-36 are rejected under 35 U.S.C. 103(a) as being unpatentable over Nazarenko et al. (US Patent 5866336) in view of Nerenberg (US Patent 6531302)

Nazarenko et al. teach a method of methylation analysis using oligonucleotides that are detectably labeled with molecular energy transfer labels which provide a rapid, sensitive, and reliable method for detecting amplification products that greatly decreases the possibility of carryover contamination with amplification products and that is adaptable to many method for amplification of nucleic acid sequences (see column 2, lines 42-52). Nazarenko et al. teach contacting a sample containing nucleic acids with bisulfite to convert unmethylated cytosines to uracil and conducting an amplification reaction in the presence of primers specific for selected target sequences (see column 32, lines 3-16). Nazarenko et al. teach a triamplificate reaction

which uses of a third oligonucleotide, blocker oligonucleotide that is ligated to the extended hairpin primer (see figure 6). Nazarenko et al. teach the two primers are complementary to the target strand and a third oligonucleotide, blocker, is partially complementary to one of two extending primers. Nazernko et al. teach the triamplificaiton utilized two thermostable enzymes, DNA polymerase and DNA ligase (claim 38 and 39). Nazarenko et al. teach during the repeated steps of polymerization and ligation, one of the extended primers is ligated to the blocker. Nazarenko et al. teach the use of hairpin primers in the amplification reaction, during the first cycle the hairpin primer will be extended and ligated to the blocker probe, during the second cycle the extended hairpin primer will become a template for the second primer (see column 26, lines 36-43). Nazarenko et al. does not teach the use of a blocker probe to prevent hybridization of probe oligonucleotides to a background DNA or detection by hybridization to oligomer arrays.

Nerenberg et al teach amplification and detection on electronically addressable microchips (see column 1, lines 9-22). Nerenberg et al. teach prevention of amplifying a background molecule by modifying the ends of the ligation probe by blocking the 3' hydroxyl group or modification of the 5' probe (results in a probe that functions as a blocking probe). Nerenberg et al. teach blocking the background by adding a probe that is modified at the 3' or 5' end that prevents hybridization and ligation of the background DNA (see column 48, lines 57-67). Additionally, Nerenberg et al. teach the use of a competitor probe that is not extendable (see column 41, lines 54-67). Nerenberg et al. teach the competitor sequence is preferably identical to nearly identical to one of the amplification primers and can compete with hybridization to the target strand (see column 42, lines 1-12).

Therefore, it would have been prima facie obvious to one of ordinary skill in the art at the time the invention was made to improve the method of detecting methylation of a target DNA using amplification by PCR as taught by Nazarenko et al. to include blocking probes and detection on a microchip as taught by Nerenberg et al. to make the method of Nazarenko et al. more versatile and efficient. The ordinary artisan would have been motivated to improve the method of analyzing the methylation pattern of the DNA by Nazarenko et al. with the method of blocking probes and array hybridization detection as taught by Nerenberg et al. because Nerenberg et al. teaches that blocking reaction can efficiently block background DNA and the microarray allows for efficient detection and optimal amplification, detection, and analysis of target nucleic acids (see column 9, lines 39-45) . The ordinary artisan would have had a reasonable expectation of success that the use of blocking probes and array analysis could be used in the method of Nazarenko et al. because both Nararenko teach ligase followed by amplification of a target nucleic acid to detect nucleic acid sequences. Furthermore, because both Nararenko and Nerenberg teach analysis of target nucleic acid by ligation and amplification using blocking probes, it would have been obvious to one skilled in the art to substitute one method, the method of ligation and amplification of PCR as taught by Nararenko, for the method of ligation and amplification of the genomic DNA sample to reduce background molecule contamination as taught by Nerenberg in order to achieve the predictable result of detecting target gene sequences.

Conclusion

13. No claims are allowable.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Sarae Bausch whose telephone number is (571) 272-2912. The examiner can normally be reached on M-F 9am-5pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ram Shukla can be reached on (571) 272-0735. The fax phone number for the organization where this application or proceeding is assigned is (571) 273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at (866) 217-9197 (toll-free).

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to (571) 272-0547.

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